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Baseline Predictors of Virological Response for Chronic Hepatitis B Patients

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Abstract

AIM: To determine which baseline factors of chronic hepatitis B patients are predictive of virological response to Peginterferon α-2b therapy.

METHODS: A total of 21 HBeAg-positive chronic hepatitis B (CHB) patients treated with Peginterferon α-2b were recruited. They were treated with Peginterferon α-2b (0.5-1.0 μg/kg per week) for 24 wk and followed up for 24 wk. Clinical and laboratory data of the patients were determined at pretreatment and at week 12, at 24 during treatment, and at week 48 during follow up.

RESULTS: Ten patients achieved a virological response at the end of treatment. Their baseline serum alanine aminotransferase (ALT), thyroid-stimulating hormone (TSH), and total thyroxin (TT4) levels were significantly different from those who failed treatment. The positive predictive values (PPV) and negative predictive values (NPV) of ALT, TSH, and TT4 were 75% and 89 %, 75% and 89 %, and 75% and 75%, respectively. Moreover, combinations of the baseline ALT and TT4, ALT and TSH, TT4 and TSH levels had much higher PPV and NPV (86% and 88%, 89% and 100%, 83% and 100%, respectively).

CONCLUSION: Baseline serum ALT, TSH, and TT4 levels, especially in combination, have high predictive values of virological response to Peginterferon α-2b in HBeAg-positive CHB patients.

Key words: Chronic hepatitis B; Hepatitis B virus; Predictors; Virological response; Peginterferon

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INTRODUCTION

Chronic hepatitis B virus (HBV) infection is a worldwide health problem. More than 400 million people are chronically infected with HBV and are at risk of developing liver cirrhosis and hepatocellular carcinoma. Each year more than one million people die from HBV-related liver diseases[1-4].

At present, the two main categories of antiviral drugs for chronic hepatitis B are interferon (including Peginterferon) and nucleoside/nucleotide analogs. Many studies have shown that an elevated serum ALT level is associated with virological response and HBeAg seroconversion in CHB patients[5-8]. Besides higher serum ALT level, some studies also showed that higher aspartate aminotransferase (AST) level, increased histological activity in biopsy specimens, female sex, and lower serum HBV DNA levels are associated with a higher probability of HBeAg seroconversion in CHB patients treated with interferon-based therapies[9-13]. It is also reported that the HBV genotype is an important predictor of response to interferon-based therapies[14-17].

Recently, serum HBeAg levels have been used as

outcome predictors of sustained virological response to Peginterferon α-2a in HBeAg-positive CHB patients and showed high negative predictive values (NPVs) at week 24 of therapy[18]. Another study showed that early serum HBsAg drops also had high predictive values of sustained virological response to Peginterferon α-2a in HBeAg-negative chronic hepatitis B patients both at week 12 and 24[19].

However, the predictive values of other factors, especially the baseline factors for virological response to Peginterferon α-2b therapy are not clear. Therefore, in this study, we aimed to determine how well the baseline factors predicted the virological response to Peginterferon α-2b therapy in HBeAg-positive CHB patients.

MATERIALS AND METHODS

Ethics

The study was approved by the Investigation and Ethics Committee for Human Research at the Peking University First Hospital (Beijing, China). All patients provided informed written consent.

Patients and study design

Twenty-one consecutive HBeAg-positive chronic hepatitis B patients were evaluated. Patients were treated with Peginterferon α-2b at a dose of 0.5-1.0 μg/kg per week for 24 wk. Clinical and laboratory data of the patients were determined before treatment, at week 12, and 24 during treatment. Thereafter they were scheduled for follow-up visits every 12 wk. End of treatment (EOT) response was defined as more than 2 $\log_{10}$ IU/mL reduction in HBV DNA levels at the EOT. Non-response was defined as less than 2 $\log_{10}$ IU/mL reduction in HBV DNA levels at the EOT.

Measurement of serologic markers of HBV

HBsAg, antibody to HBsAg, HBeAg, antibody to HBeAg and anti-HBc were measured using a microparticle enzyme immunoassay (Abbott Laboratories, North Chicago, IL). The HBV genotype was determined using the INNO LiPA HBV genotyping assay. Serum HBV DNA was measured using the TaqMan polymerase chain reaction assay [COBAS TaqMan, Roche Molecular System (lower limit of detection, 20 IU/mL)].

Measurement of biochemical markers

Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were measured using a Hitachi Model 7600 Series Automatic Analyzer (Hitachi). Thyroid-stimulating hormone (TSH), total triiodothyronine (TT3), and total thyroxin (TT4) levels were measured using a Centaur Automated Chemiluminescence System (Bayer).

Statistical analysis

Quantitative variables were expressed as the median with interquartile ranges (IQR), and categorical variables as frequencies. Comparisons between groups of quantitative and qualitative variables were performed using the Mann-Whitney *U* test and the Fisher's exact test, respectively. The accuracy of serum factors to predict virological response was assessed using the receiver operating characteristic curve. The cutoff value was chosen according to the receiver operating characteristic curve when the sensitivity and specificity were both relatively high for the selective baseline factor. All tests were two-sided and used a significance level of 0.05. Data handling and analysis were performed with SPSS software for windows, version 13.0 (SPSS Inc., Chicago, IL).

RESULTS

Baseline characteristics of patients

The baseline characteristics of the 21 HBeAg-positive CHB patients are shown in Table 1. The median age was 25 years (range, 20-39), and 81% of them were male (17/21). The median value of serum HBV DNA levels was 8.2 $\log_{10}$ IU/mL (IQR, 7.5-8.7 $\log_{10}$ IU/mL). The distribution of HBV genotype was: B, 24%; C, 76%. The median values of serum ALT, AST, TSH, TT3, and TT4 level were 147 IU/L (IQR, 123-201 IU/L), 65 IU/L (IQR, 51-97 IU/L), 2.06 mIU/L (IQR, 1.41-3.10 mIU/L), 2.22 nmol/L (IQR, 2.04-3.03 nmol/L), and 111.4 nmol/L (IQR, 96.8-140.6 nmol/L) respectively. The baseline TT3 and TT4 values of one patient were not assayed at pretreatment. Serological tests were negative for hepatitis C virus, hepatitis D virus, and human immunodeficiency virus in all patients.

Virological response

Of the 21 patients, ten (48%) showed an EOT response, and eleven (52%) were non-responders. Four patients (19%) obtained HBeAg seroconversion at the end of treatment (week 24). However, two of the four HBeAg seroconversion patients lost anti-Hbe, while another six patients achieved HBeAg seroconversion at week 48. The median value of serum HBV DNA levels were 2.7 $\log_{10}$ IU/mL (IQR, 1.9-4.0 $\log_{10}$ IU/mL) and 3.1 $\log_{10}$ IU/mL (IQR, 1.8-6.6 $\log_{10}$ IU/mL) in responders at week 24 and 48 respectively. In non-responders, The median value of serum HBV DNA levels were 7.4 $\log_{10}$ IU/mL (IQR, 6.8-7.9 $\log_{10}$ IU/mL) and 7.6 $\log_{10}$ IU/mL (IQR, 7.1-8.7 $\log_{10}$ IU/mL) at week 24 and 48 respectively. The baseline ALT and TT4 level were significantly higher in responders than in non-responders(both $P < 0.05$, Table 1). However, the baseline TSH level was significantly lower in responders than in non-responders($P < 0.05$, Table 1). The baseline age was similar between responders and non-responders.

Predictability

To determine how well the baseline ALT, TSH and TT4 levels predicted virological response to Peginterferon α-2b therapy, we performed receiver operating characteristic curves for each parameter. The areas under the curves of

Table 1 Baseline characteristics of patients

Characteristic	All patients (n = 21)	Responders (n = 10)	Non-responders (n = 11)	P value
Median age, range (yr)	25 (20-39)	25 (20-38)	25 (20-39)	0.749
Gender, male (%)	81	70	91	0.311
HBV genotype (%B, C)	24, 76	10, 90	36, 64	0.311
Median HBV DNA levels, range [log (IU/mL)]	8.2 (7.5-8.7)	7.7 (7.2-8.4)	8.4 (8.1-8.8)	0.090
Median ALT level, range (IU/L)	147 (123-201)	184 (146-247)	124 (112-148)	0.011[a]
Median AST level, range (IU/L)	65 (51-97)	90 (57-132)	64 (45-73)	0.072
Median TSH levels, range (mIU/L)	2.06 (1.41-3.10)	1.82 (1.14-2.08)	2.55 (1.68-4.11)	0.035[c]
Median TT3 levels, range (nmol/L)	2.22 (2.04-3.03)	2.85 (2.02-3.85)	2.20 (2.02-2.54)	0.305
Median TT4 levels, range (nmol/L)	111.4 (96.8-140.6)	132.7 (109.0-168.5)	107.8 (88.4-117.3)	0.037[e]

Data are expressed as the median (IQR) and as percentages. HBV: Hepatitis B virus; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; TSH: Thyroid-stimulating hormone. [a]P < 0.05 differences of baseline serum ALT level between responders and non-responders; [c]P < 0.05 differences of baseline serum TSH level between responders and non-responders; [e]P < 0.05 differences of baseline serum TT4 level between responders and non-responders.

Table 2 Predictive value of single and combined baseline factors

Parameters	Responders	Non-responders	Predictive value (%)
ALT ⩾ 140	9	3	PPV = 75
ALT < 140	1	8	NPV = 89
TSH < 2.4	9	3	PPV = 75
TSH ⩾ 2.4	1	8	NPV = 89
TT4 ⩾ 120	6	2	PPV = 75
TT4 < 120	3	9	NPV = 75
ALT ⩾ 140, TT4 ⩾ 120	6	1	PPV = 86
ALT < 140, TT4 < 120	1	7	NPV = 88
ALT ⩾ 140, TSH < 2.4	8	1	PPV = 89
ALT < 140, TSH ⩾ 2.4	0	6	NPV = 100
TT4 ⩾ 120, TSH < 2.4	5	1	PPV = 83
TT4 < 120, TSH ⩾ 2.4	0	7	NPV = 100

Data are expressed as numbers of instances. PPV: Positive predictive values; NPV: Negative predictive values.

ALT, TSH, and TT4 were 0.827 (P = 0.011), 0.773 (P = 0.035), and 0.778 (P = 0.037), respectively. Accordingly, we chose cutoff values of 140 IU/L, 2.4 mIU/L, and 120 nmol/L for ALT, TSH, and TT4, respectively. Correspondingly, their positive predictive values (PPV) and negative predictive values (NPV) were 75% and 89 %, 75% and 89 %, and 75% and 75% (Table 2). We further performed the combination of the baseline ALT and TT4, ALT and TSH, and TT4 and TSH to predict the virological response. We found that their PPV and NPV were 86% and 88%, 89% and 100%, and 83% and 100%, respectively (Table 2).

DISCUSSION

Nowadays, more and more doctors are taking the initiative in individualized treatment for chronic hepatitis B patients. With the purpose of taking individualized treatment, it is important to evaluate the baseline status of each patient at the start of treatment and to then decide which antiviral drug is the best choice. For those patients who are not likely to benefit from Peginterferon α-2b therapy, an early switch to nucleoside/nucleotide analogs is essential.

Recently, a study showed that HBeAg levels had high negative predictive values (NPVs) at week 24 of sustained virological response to Peginterferon α-2a in HBeAg-positive CHB patients[18]. While in HBeAg-negative CHB patients, early serum HBsAg drops also had high predictive values of sustained virological response to Peginterferon α-2a at week 12 and 24[19].

In our study, 21 HBeAg-positive CHB patients were treated with Peginterferon α-2b for 24 wk and followed up for 24 wk. We found that baseline serum ALT, TSH, and TT4 levels, and especially the combination of these factors, had high predictive values of virological response to Peginterferon α-2b therapy.

To identify the baseline predictors of virological response, we performed univariate analysis and receiver operating characteristic curves for baseline serum ALT, TSH, and TT4 levels, and found that the cutoff value of 140 IU/L of baseline serum ALT level had a relatively high predictive value of virological response. The cutoff values for TSH and TT4 were 2.4 (mIU/L) and 120 (nmoI/L), respectively. Moreover, we found that combinations of these factors could further improve the PPV and NPV scores.

Some studies have shown that the rates of HBeAg loss and seroconversion were correlated with the baseline level of ALT. In patients with a higher baseline level of ALT, the rates of HBeAg loss and seroconversion during lamivudine therapy were also significantly higher at the end of year three[7]. A previous study showed that CHB patients with normal ALT levels respond very poorly to interferon α-2a therapy. However, the response was significantly better in patients with elevated ALT levels[13]. In HBeAg-negative CHB patients treated with Peginterferon α-2a, with or without lamivudine, a high baseline ALT level was identified as a significant predictor of virological response at weeks 24 post-treatment[8].

Besides high baseline serum ALT level, we also found that higher TT4 level and lower baseline serum TSH level were associated with better outcome of Peginterferon α-2b therapy in HBeAg-positive CHB patients.

Although no study exploring the predictive value of virological response for baseline serum TT4 in chronic hepatitis B patients has been reported, several studies have demonstrated a reciprocal relationship between the endocrine and immune systems. Recently a study showed that triiodothyronine and thyroxin concentrations were

positively associated with markers of inflammation, natural killer-like T cells, activated monocytes derived interleukin-6 (IL-6), higher expression of IL-2 receptor on CD3+ T-lymphocytes, and percentage expression of memory T-lymphocytes, memory T-helper lymphocytes and memory T-cytotoxic lymphocytes within normal physiological ranges[20]. This is supported by previous findings that thyroid hormone was involved in primary and secondary lymphopoiesis, and blastogenic responses to T and B cell mitogens were also enhanced following thyroxin administration[21,22]. Other studies showed that thyroxin did not induce resting T lymphocyte proliferation but increased mitogen ConA-induced stimulation after three days of culture, in a dose-dependent manner. Thyroxin substitutive treatment restored the euthyroid status and reversed the impairment of T-cell activation induced by chronic stress in mice[23,24]. Interestingly, the age-dependent immunological deterioration in old mice could be recovered by thyroxin treatment[25]. These results indicated that thyroxin could enhance the immune response. Thus, this may be the reason why the responders who had higher baseline TT4 level achieved virological response more easily during Peginterferon α-2b therapy in our study.

Another major finding was the lower baseline TSH level of responders was also associated with higher virological response rate. This could be caused by the negative feedback mechanism due to their higher baseline serum TT4 level.

In conclusion, the identification and application of baseline factors to predict virological response of chronic hepatitis B patients before antiviral therapy is important. Using this method, we can identify patients who will most likely benefit from Peginterferon α-2b therapy before treatment. However, because of the small cohort of patients enrolled in our study, large-scale studies are needed to further confirm our results and to identify simpler and more appropriate factors that have high predictive values of virological response in chronic hepatitis B patients.

COMMENTS

Background

Early prediction of virological response for chronic hepatitis B patients treated with antiviral drugs is important. Some factors such as HBsAg and HBeAg reduction have been found to have high predictive values of sustained virological response in chronic hepatitis B patients treated with Peginterferon α-2a. However, the predictive values of other factors, especially the baseline factors for virological response to Peginterferon α-2b therapy, are not clear.

Research frontiers

Many studies have shown that an elevated serum alanine aminotransferase (ALT) level was associated with virological response and HBeAg seroconversion in CHB patients. Recent studies showed a reciprocal relationship between the endocrine and immune system. In this study, the authors showed that baseline serum ALT, thyroid-stimulating hormone (TSH), and total thyroxin (TT4) levels, and especially combinations of these factors, have high predictive values of virological response to Peginterferon α-2b in HBeAg-positive CHB patients.

Innovations and breakthroughs

The present study demonstrated that baseline serum ALT, TSH, and TT4 levels, and especially combinations of these factors, have high predictive values of virological response to Peginterferon α-2b in HBeAg-positive chronic hepatitis B (CHB) patients before treatment.

Applications

This study might represent a future strategy for identifying chronic hepatitis B patients who will most likely benefit from Peginterferon α-2b therapy before treatment.

Terminology

ALT is an enzyme that is normally present in liver and heart cells. ALT is released into blood when the liver or heart is damaged. TSH is a peptide hormone synthesized and secreted by thyrotrope cells in the anterior pituitary gland which regulates the endocrine function of the thyroid gland. Thyroxin (T4) is a form of thyroid hormone which is the major hormone secreted by the follicular cells of the thyroid gland.

Peer review

This study is of interest as it describes the relationship of virological response to Peginterferon α-2b therapy and serum parameters at pretreatment, although this was obtained in a very small cohort of patients.

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